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THE ACTINIC AND VISUAL FOCUS IN MICRO-PHOTOGRAPHY WITH HIGH POWERS.

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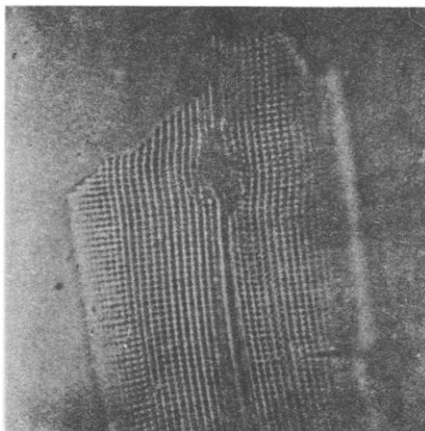
[PLATE I.]

We find it commonly said that whilst the difference between the visual and the actinic focus is considerable when making micro-photographs with low powers, it is not appreciable when using high powers. My experience does not accord with this statement, and some notes upon my own experiments may have interest to others.

If the statement had been that a *sharp picture* may be taken when the object is exactly in focus with a high power, I should not take exception to it, and I incline to think that this is what has been meant. But a sharp picture may be either a positive or a negative of the visual image seen in the microscope, and in my own work so many examples have turned out to be positives when I expected them to be negatives, that I have been led to make an investigation of the subject, in which the evidence tends strongly to show that with our best high power lenses the image fixed upon the sensitized plate is a positive instead of being a negative, and consequently the paper prints from this are negatives and not positives.

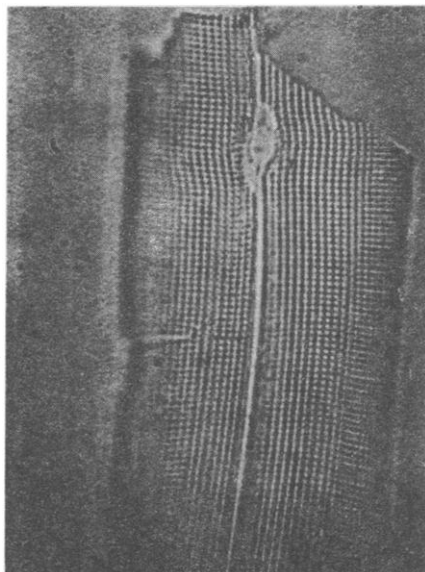
It would be very easy to overlook this difference in a large class of micro-photographs, because, in an alternation of dark and light lines, or dark and light spaces, it often matters little which of a pair is light or dark; the picture may be equally clear and satisfactory either way. In the case of a large majority of the microscopic objects photographed, either the positive or negative image would be good enough for the purpose intended: so good that a close examination of the point I am now suggesting would hardly occur to one. This, in fact, was my own experience until, in efforts to get a good picture of the broken edge of fragments of the finer diatoms, my attention was arrested by the fact that the appearances seen by

PLATE I.



EXPLANATION OF PLATE I.

The figure in the smaller rectangle is printed from a true *negative* of *Pleurosigma balticum*; that in the larger rectangle is printed from the *positive*, as explained in the foregoing paper.



the eye were often reversed in the print from the supposed negative which I had taken. As, in dealing with minute areolæ this often amounted to showing a projection where I had seen an apparent depression, and *vice versa*, it became in effect a failure to photograph what I had seen, and challenged my best efforts to overcome the difficulty. If the illumination of such transparent objects as diatoms were always by a perfectly central beam of parallel rays of light, there would be no practical difference whether they showed light upon a dark ground, or the reverse. But we rarely get such exactly central illumination, even after our best efforts to do so. For example, plate No. 23 of my broken shell series was thus taken with light intended to be strictly central, a diaphragm being behind the achromatic condenser, which had a small circular hole in it, limiting the illuminating rays to the small central portion of the condenser. Yet in one position the central areolæ of the *Coscinodiscus* which it represents, appear as deep cups, whilst, if the photograph be turned around so as to change places of top and bottom, they appear as projecting bosses.

No. 51 of the same series was the first in which I distinctly marked in my note-book the fact that the dots in that diatom, *Mastogloia angulata*, appeared dark in the instrument, but light in the photograph print. The difference of effect was least important in shells which are an even, smooth film of comparatively little thickness, and greatest in those in which the diatom seems to have strongly marked bars separating the lines of areolæ, as in *Pleurosigma balticum*.

In a number of cases in which the plates were originally taken with a sharp focus upon the view of the shell which I desired, I have taken transparencies from them by contact, and using these last as negatives from which to print the paper prints, I have found that these last are, according to my notes, what the former should have been if there were no difference between the visual and the actinic focus. A few of these have been prepared for exhibition to the Society. The prints taken from the second plates are marked "positives" of the originals, and are in fact the true representation of the object as I saw it when taking the original photograph. They are No. 66, *Navicula serians*, Kutz'g, taken with a Spencer $\frac{1}{8}$

objective, balsam angle 125° , with No. 118 as the "positive" from it.

No. 60, *Pleurosigma formosum*, W. Sm., taken with a Spencer $\frac{1}{8}$ objective, balsam angle 108° , with No. 122 as the "positive" from it.

No. 83, *Pleurosigma formosum*, W. Sm., taken with a Wales $\frac{1}{8}$ objective, balsam angle 82° , with No. 119 as the "positive" from it.

No. 110, *Pleurosigma balticum*, W. Sm., taken with a Zeiss $\frac{1}{8}$ objective, balsam angle 116° , with No. 113 as the "positive" from it.

The accompanying plate gives a reproduction of the last pair.

The objectives are all of the first class, and it is safe to assume that what holds true with them will be found true with any of our best glasses.

In taking the original photographs, I used a plain plate of glass instead of the usual ground glass screen in the camera, and focussed by the aid of a Dorlot focussing glass.

The examples to which I have referred would seem to warrant the conclusion that in using high power objectives the difference between the visual and the actinic focus is the equivalent of that between a positive and negative image of the object when the details have passed a certain limit in fineness. But some experiments, made for the purpose of finding how far the tube of the microscope must be moved to secure the proper actinic focus upon the sensitive plate, have had such unsatisfactory results as make me unwilling to venture any positive conclusion, but content myself with stating the facts above given, until further investigations which I am making shall be completed.

In the course of the experiments referred to, I noticed that the image taken on the plate was apparently of a lower plane in the object than the visual one which I was seeking to get. This was shown in the diatoms with a convex surface, by the sharper image, in the print or plate, of areolæ nearer the margin of the object than those upon which I had focussed. It showed also that the difference seemed to be the same in kind as in the use of low power objectives, with which it is necessary to raise (withdraw) the tube after getting a sharp visual image of the object. Acting upon this, I tried in several instances the gradual raising of the tube, taking pic-

tures at slightly varying departures from the visual focus, until the image was quite spoiled and blurred to the eye. I made some series of as many as five or six plates, thus progressively varying, but without satisfactorily establishing any point (different from the visual focus) at which the objective should be placed to secure in the photographic image the true characters of the visual one. I was surprised to find at what a distance from the visual focus *a* sharp image could be taken, but it was not *the* image for which I was in search. Examples of this sort are among the prints which I will exhibit to the Society.

I design to add to my experiments on the subject the examination of the effect of changing the focus of the focussing glass to correspond with the difference between the visual image of a diatom showing light dots or areolæ and that which shows dark ones. Everybody has noticed that a slight change of focus with a high power produces this change of appearance, and if the focussing glass were adjusted for the image which is complementary to the one desired and then the focussing done in the usual way, the result might be that which is sought. It has at least seemed worth the experiment, but a press of other work has prevented my making a satisfactory test of it before the time of our meeting.